JC12 Rec'd PCT/PTC 21 OCT 2005

Amended Claims

1. A device for contamination free preparation of analyte containing sample solution (P), comprising

5

15

- a first (2) and second chamber (3), which are connected by a channel (6, 8, 10),
- wherein the first chamber (2) has means (4) for reversibly changing its volume, and the second chamber (3) has a reversibly changeable volume,
 - wherein a connector (7, 9), which is provided with a means of flow regulation, is connected to the channel (6, 8, 10) or one of the chambers (2, 3) for loading of a sample solution into the first (2) or the second chamber (3),

characterized in that

- 20 there is provided at least one further chamber (15, 20, 21) being filled with a reactant and sealed, the further chamber being connectable to the channel (6, 8, 10) prior to use.
- 2. The device of claim 1, wherein the chambers (2, 3) and the channel (6, 8, 10) are designed as a single use device.
 - 3. The device of claims 1 or 2, wherein no means of flow regulation is provided between the first (2) and the second chamber (3).

30

4. The device of any one of claims 1 to 3, wherein the second chamber is provided with a means for reversibly changing the volume.

10-09-2005 EP0404125

5. The device of any one of claims 1 to 4, wherein a first connector (7) is connected either to a first channel (6), which extends from an end of the first chamber (2) opposite the first means (4) for reversibly changing the volume of the first chamber (2) or to the first chamber (2).

- 6. The device of any one of claims 1 to 5, wherein a second connector (9) is connected to a second channel (8), which extends from an end of the second chamber opposite the second means (5) for reversibly changing the volume or to the second chamber (3).
- 7. The device of any one of claims 1 to 6, wherein a second channel (8) extends from an end of the second cylinder opposite the second piston (5), wherein the channel (8) is connected to the first channel (6) or the first chamber (2).
- 8. The device of any one of claims 1 to 7, wherein the further chamber (15, 20, 21) has a reversibly changeable volume, preferably a means for reversibly changing the volume and wherein that chamber(s) is (are) connected with the first (2) and/or second chamber (3) through the first (6) or second channel (8) or through a further channel (s) (17, 24, 25), which is (are) connected to a third channel (10) connecting the first (6) and second channel (8).
 - 9. The device of any one of claims 1 to 8, wherein a means of flow regulation is provided between the first (2) and the second chamber (3) on one side and at least one further chamber(s) (15, 20, 21) on the other side.
 - 10. The device of any one of claims 1 to 9, wherein at least one channel between two of the further chambers (15, 20, 21) has a volume larger than the total compressible volume of the system.

5

10

15

30

- 11. The device of any one of claims 1 to 10, wherein at least one of the chambers are conically tapered at the end of the chamber opposite the means (4, 5, 14, 22, 23) for reversibly changing the volume with the opening of the respective channels (6, 8, 17, 24, 25) located at the tip of the conus.
- 12. The device of any one of claims 1 to 11, wherein the means for changing the volume in the first (2), second (3) and/or further chamber(s) (15, 20, 21) is a piston (4, 5, 14, 22, 23).
- 13. The device of claim 12, wherein the piston(s) have the shape of the end of the chamber opposite to them or can accommodate this shape.
- 14. The device of claims 12 or 13, wherein the pistons (4, 5) of the first (2) and/or the second chamber (3) comprise an elastic material, which has or can accommodate the shape of the end of the chamber opposite to them.
 - 15. The device of any one of claims 12 to 14, wherein the pistons (16, 22, 23) in further chambers (15, 20, 21) comprise a reduced elasticity in comparison to the pistons (4, 5) in the first chamber (2) and/or second chamber (3).
 - 16. The device of any one of claims 12 to 15, wherein the pistons (16, 22, 23) are not connected to a piston rod.
- 17. The device of any one of claims 1 to 16, wherein the 30 chambers (2, 3, 15, 20, 21) have an essentially round cross-section.
 - 18. The device of any one of claims 1 to 17, wherein at least one chamber (2, 3, 15, 20, 21) preferably at least the second

5

10

15

20

- (3) and/or the further chamber(s) (15, 20, 21) are connectible to the channel(s) (8, 17, 24, 25).
- 19. The device of any one of claims 1 to 18, wherein the axes of the chambers (2, 3, 15, 20, 21) are arranged parallel to each other.
 - 20. The device of any one of claims 1 to 19, wherein in one of the chambers (2, 3, 15, 20, 21), preferably in the second chamber (3) a liquid (L) is provided capable of solubilizing organic substances comprising an analyte and wherein the organic substances are preferably cells.
- 21. The device of any one of claims 1 to 20, wherein in at least one of the chambers (2, 3, 15, 20, 21) or in at least one of the channel(s) (6, 8, 10, 17, 24, 25) magnetic particles (18) are provided capable of binding the analyte.
- 22. The device of claim 21, wherein the magnetic particles 20 (18) have a diameter in the range from 50 nm to 50 μ m, preferably from 200 nm to 20 μ m.
- 23. The device of any one of claims 1 to 22, wherein in one of the chambers (2, 3, 15, 20, 21), preferably in a further chamber (20), a wash solution (W) is provided.
 - 24. The device of any one of claims 1 to 23, wherein in one of the chambers (2, 3, 15, 20, 21), preferably in a further chamber (21), an elution solution (E) is provided.
 - 25. The device of any one of claims 1 to 24, wherein the connectors (7, 9) are provided with a means of flow regulation, preferably a valve or septum.

10

IQ-09-2005 EP0404125

26. The device of any one of claims 1 to 25, wherein the chambers (2, 3, 15, 20, 21) are fluid tight against the surrounding when the connector(s) (7, 9) are closed.

- 5 27. The device of any one of claims 1 to 26, equipped to accommodate the positioning of a magnet at the end of the chamber(s) (2, 3, 15, 20, 21), preferably the first (2) or the second chamber (3).
- 10 28. The device of any one of claims 1 to 27, wherein the device is provided with an enclosure (1) and wherein the enclosure (1) is preferably made of synthetic material.
- 29. The device of any one of claims 1 to 28 wherein the chan-15 nels (6, 8, 17, 24, 25) and the connectors (7, 9) are comprised in a base plate (30).
- 30. The device of claim 29, wherein at least the first chamber, preferably all chambers (2, 3, 15, 20, 21) open up towards the edge of the enclosure (1), so that the means (4, 5, 14, 22, 23) for reversibly changing the volume can be operated from the outside.
- 31. The device of claims 29 or 30, wherein the enclosure (1) and/or the base plate (30) provided with a means (13) for attaching the device in a corresponding receptacle to allow automatic changing of the volume of at least one chamber (2, 3, 15, 20, 21).
- 32. A kit of parts comprising a base plate comprising a channel (6, 8, 10), wherein a connector (7, 9), which is provided with a means of flow regulation, is connected to the channel (6, 8, 10) and at least one at least one chamber (15, 20, 21) being filled with a reactant and sealed, the chamber (15, 20, 21) being connectable to the channel (6, 8, 10) prior to use.

EP0404125

5

15

- 33. A method for contamination free preparation of analyte(s) from organic substance comprised in a sample solution (P), using a device according to any one of claims 1 to 31 comprising the following steps:
- introducing a predetermined volume of sample solution (P) through a connector (7, 9) into the first (2) or second chamber (3),
- 10 interrupting the flow directed through the connector (7, 9),

moving back and forth of the sample solution (P) between the first (2) and the second (3) chamber in such that the sample solution is contacted with material binding or adsorbing the analyte and that the analyte(s) comprised in the sample solution (P) can bind or adsorb to the material, and

dislodging of the analyte(s) through a connector (7, 9).

- 20 34. The method of claim 33 comprising the following step: optionally eluting the analyte from said material.
- 35. The method of claim 33 or 34, wherein the analyte is selected from the group consisting of nucleic acids and polypeptides.
- 36. The method of one of claims 33 to 35, wherein the material binding or adsorbing the analyte coats at least part of the surface of the chambers (2, 3, 15, 20, 21) and/or channels (6, 8, 17, 24, 25) or particles, in particular magnetic particles comprised within the chambers and/or channels.
- 37. The method of claim 36, wherein the analyte(s) is (are) dislodged bound to the magnetic particles (18) or separate from the magnetic particles (18).

0-09-2005 EP0404125

38. The method of one of claims 33 to 37, wherein the sample solution (P) is moved back and forth by alternately extending and reducing the volume of the first chamber (2).

- 5 39. The method of one of claims 33 to 38, wherein the sample solution (P) is moved back and forth by alternately moving the first (4) and the second means (5) for reversibly changing the volume towards the end of the chamber opposite to the means (4, 5).
- 40. The method of one of the claims 33 to 39, wherein the sample solution (P) is in a further step sonicated and/or mixed with a liquid (L) for solubilization of the organic substance comprised in the sample solution (P).
- 41. The method of one of the claims 36 to 40, wherein the magnetic particles (18) and the analyte bound thereon are retained in a predetermined region of a chamber (2, 3), preferably the end of the first (2) or second chamber (3) opposite to the means for reversibly changing the volume (4, 5) by generating a magnetic field in the predetermined region of the chamber (2, 3).
- 42. The method of one of claims 33 to 41, wherein in a fur-25 ther step sample solution (P) depleted of analyte(s) is substantially removed from the chamber(s) (2, 3).
- 43. The method of claims 33 or 42, wherein a wash solution
 (W) provided in one further chamber (20) is flown over surface
 30 coated with binding material or mixed with the particles,
 preferably magnetic particles (18).
 - 44. The method of claim 43, wherein the magnetic particles (18) and the analyte bound thereon are retained in a predetermined region of a chamber, preferably the end of the first (2) or second chamber (3) opposite to the means for reversibly

10

15

EP0404125

changing the volume (4, 5) by generating a magnetic field in the predetermined region of the chamber (2,3).

- 45. The method of claims 43 or 44, wherein in a further step wash solution (W) is substantially removed from the chamber(s) (2, 3).
- 46. The method of one of claims 33 to 45, wherein an elution solution (E) provided in one further chamber (21) flown over surface coated with binding material or mixed with the particles, preferably magnetic particles (18).
- 47. A method according to claim 46, wherein the magnetic particles (18) are retained in a predetermined region of a chamber, preferably the end of the first (2) or second chamber (3) opposite to the means for reversibly changing the volume (4, 5) by generating a magnetic field in the predetermined region of the chamber.
- 20 48. The method of claims 46 or 47, wherein in a further step elution solution (E) comprising the analyte is substantially dislodged from the chamber (2, 3).
- 49. The method of one of claims 46 to 48, wherein the elution volume is in a range from about 1 to about 100 μ l.
 - 50. The method of one of claims 33 to 49, wherein the sample solution (P) depleted of analyte, the liquid (L) for solubilization mixed with the sample solution (P) depleted of analyte, the wash solution (W) and/or the magnetic particles (18) are collected in one of the chambers (2, 3, 15) and are discarded after dislodging, preferably together with the enclosure (1).
- 51. The method of one of claims 33 to 50, wherein the flow of liquids between two chambers (2, 3, 15, 20, 21) is controlled

EP0404125

by alternately extending and reducing the volume of one chamber (2, 3, 15, 20, 21), while keeping the volume of all but one of the other chambers (2, 3, 15, 20, 21) constant.

- 5 52. The method of one of claims 33 to 50, wherein the flow of liquids between two chambers (2, 3, 15, 20, 21) is controlled by alternately moving one means (4, 5, 14, 22, 23) and a second means (4, 5, 14, 22, 23) for reversibly changing the volume towards the end of the chamber (2, 3, 15, 20, 21) opposite to the means, while keeping the volume of all other chambers (2, 3, 15, 20, 21) constant.
- 53. A method according to one of the claims 33 to 52, wherein no further liquid except the sample solution (P) is introduced into the system formed by the chambers (2, 3, 15, 20, 21) and the channels (6, 8, 10, 17, 24, 25).